Subacute meningoencephalitis in a subset of patients with AD after Aβ42 immunization

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Abstract—*Background:* AD is characterized by cerebral deposition of β-amyloid plaques with amyloid β-peptide (Aβ) 42 as the major peptide constituent, along with neurofibrillary tangles and neuronal loss. In transgenic mice, active immunization against Aβ42 removes these plaques and improves cognitive function. A Phase I study in AD patients demonstrated good safety and tolerability of multiple injections of aggregated Aβ42 (AN1792) with QS-21 as adjuvant. *Methods:* Three hundred seventy-two patients with mild to moderate AD were randomized to receive IM injections of AN1792 or placebo (4:1) at baseline and at months 1, 3, 6, 9, and 12 in a multicenter Phase II safety, tolerability, and pilot efficacy study. Dosing was terminated after four early reports of meningoencephalitis, but follow-up continued. The study remains blinded, and further results will be reported after its termination. *Results:* Symptoms and laboratory findings consistent with meningoencephalitis occurred in 18 of 298 (6%) patients treated with AN1792 compared with 0 of 74 on placebo ($p = 0.020$). Sixteen of the 18 had received two doses, one had received one dose, and one had received three doses of the study drug before symptoms occurred. The median latency from the first and last injections to symptoms was 75 and 40 days. No case occurred later than 6 months after the first immunization. Anti-Aβ42 antibody titers were not correlated with the occurrence or severity of symptoms or relapses. Twelve patients recovered to or close to baseline within weeks, whereas six remain with disabling cognitive or neurologic sequelae. All 18 patients remain alive to date (December 31, 2002), 6 months to >1 year after symptom onset. *Conclusions:* Postvaccination meningoencephalitis occurred without clear relation to serum anti-Aβ42 antibody titers. Potential mechanisms such as T-cell and microglial activation may be responsible and are under consideration to develop a safer anti-Aβ immunotherapy for AD.

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AD is characterized by progressive memory loss, cognitive decline, and loss of functional abilities, ultimately leading to complete dependency and death. It affects approximately 30 million people worldwide.[1] Although acetylcholinesterase inhibitors are efficient as symptomatic therapy for AD, they do not stop or reverse disease progression, so there is currently no cure for this devastating illness. Much evidence supports the central role of the amyloid β-peptide (Aβ) 42 in the pathogenesis of AD.[2,3] Thus, treatment strategies aimed at reducing the formation or promoting the clearance of Aβ42 are being developed with the aim of halting the progression of the disease process.

Active immunization against preaggregated Aβ42 effectively reduced the amount of β-amyloid plaques in the brains of transgenic mice expressing the mutations implicated in dominantly inherited AD.[4,5] After immunization with Aβ42, a reduction in Aβ neuropathology[4,6,7] and improvements in cognitive performance were reported, with[8,9] or without[10] a reduction of the amyloid deposits in the brain. Based on

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The co-authors of this article are the study investigators who reported at least one case of meningoencephalitis and agreed to collaborate on the manuscript. J.-M.O. is a scientific adviser to the sponsors, and S.G. chairs the Safety Monitoring Committee for this trial.

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these preclinical findings and after extensive preclinical safety studies in several species, immunization with preaggregated synthetic Aβ42 (AN1792) combined with the immunogenic adjuvant QS-21 was tested in Phase I studies which demonstrated good safety and tolerability in 104 AD patients treated with single or multiple doses of AN1792 or with QS-21 alone and elicited a detectable rise of anti-Aβ42 antibodies in about 25% of patients who received AN1792.[11] Thus, an international, multicenter Phase II study of active immunotherapy with Aβ42 (AN1792) plus QS-21 as adjuvant was initiated in 2001 to evaluate its safety, tolerability, and pilot clinical efficacy.

Dosing was terminated in January 2002 after signs, symptoms, and laboratory findings consistent with meningoencephalitis were reported in four patients treated with AN1792.[12] Thorough clinical follow-up and monitoring of the nonaffected patients are continuing under blinded conditions. This article provides an overview of the design of the Phase II study and describes in detail or summarizes in tables the case histories of patients who developed manifestations of meningoencephalitis, including clinical signs and symptoms, CSF abnormalities, serum antibody titers, MRI findings, and outcome.

Patients and methods. *Patients.* Eligible patients were aged 50 to 85 years and met the clinical criteria for a diagnosis of probable AD according to National Institute of Neurological and Communication Disorders and Stroke/Alzheimer's Disease and Related Disorders Association criteria.[13] A baseline MRI was required to support the diagnosis of AD and to exclude other structural causes of dementia. Additional criteria included a score of 15 to 26 on the Mini-Mental State Examination (MMSE),[14] a Rosen modified ischemic score of ≤4 to exclude vascular dementia, and written, informed consent from the patient and patient's caregiver. Nonprescription or prescription medications other than acetylcholinesterase inhibitors for cognitive enhancement were not permitted in the 3-month period prior to inclusion. The study was carried out according to the Declaration of Helsinki and subsequent revisions and was approved by appropriate local or national ethics committees.

Study design. This multicenter, randomized, parallel, double-blind, placebo-controlled trial was undertaken in 30 centers in the USA and Europe. It was scheduled to run for 2 years, including screening, enrollment, follow-up, and analysis. A total of 372 patients with mild to moderate AD were randomized to receive active treatment or placebo in a 4:1 ratio, with the aim of achieving a final 1:1 comparison of patients with an AN1792 antibody response vs placebo.

The planned immunization schedule involved injections in the deltoid muscle (0.5 mL) of 225 μg AN1792 (synthetic, preaggregated Aβ42) combined with 50 μg QS-21 (immunogenic adjuvant) or saline placebo (saline solution without adjuvant) at baseline and at months 1, 3, 6, 9, and 12.

The primary outcome measures were safety and tolerability, the AD Assessment Scale—Cognitive to assess changes in memory and other cognitive functions, and MRI scanning to evaluate change in whole-brain volume. Immunogenicity was assessed by ELISA analysis of serum samples for anti-AN1792 antibody titers at baseline and at monthly intervals thereafter. CSF levels of anti-AN1792, tau, and Aβ were also evaluated at baseline and planned for evaluation at the end of the trial. Safety monitoring throughout the study was assessed by adverse event (AE) reporting, physical and neurologic examinations, vital signs, and standard laboratory evaluations.

Statistical methods. After suspension of active dosing because of the first reports of serious CNS AE described herein, the blind was broken for the serious AE (SAE) cases. As the study remains blinded for the non-SAE cases, the statistics for this report are limited to 1) comparisons of the probability of occurrence of these SAE cases in the actively immunized vs placebo group and comparisons within groups with the Fisher's exact test, two tailed; and 2) comparisons of the frequency of occurrence with the χ^2 test between Europe and the USA and between France, other European countries, and the USA.

Results. Enrollment of patients began in October 2001 and continued through December 2001. Active dosing of the study drug was suspended by the Safety Monitoring Committee on January 11, 2002, after the first reports of four patients who developed signs and symptoms consistent with aseptic meningoencephalitis during the preceding 3 weeks. At that time, most of the 372 patients had received two injections (338), whereas 4 had only one injection and 30 had received three injections. All SAE cases were included in the investigational new drug safety reports. To date (December 31, 2002), 18 patients have been withdrawn from the study because they developed meningoencephalitis (table 1). The latency period from the last injection to symptom onset varied from 5 to 71 days (median 40 days) with two outliers at 156 and 168 days. The time from the first injection to onset of symptoms varied from 16 to about 100 days, excluding the two outliers (median 75 days). The most delayed case (Patient 18) was retrospectively identified as having occurred 195 days after the first injection. No case was reported >6 months after the last injection during the 1-year scheduled follow-up (figure 1). The blind was broken for these patients, and all had received AN1792. Blinding stays unbroken for most of the other 355 patients, including some patients who have elected to stop participation in the study. Monitoring of patients in the study continued for 12 months after the baseline evaluation until the last scheduled follow-up, and the last protocol-scheduled follow-up visit was on December 17, 2002. As the study remains blinded, no definitive statement can be made about injection site reactions and whether they unblinded to the investigators: Mild to moderate pain occurred in some patients in the Phase I 102 study, but as all raters in the current study were blinded to AE data, it is unlikely that these local reactions may have biased the diagnosis of meningoencephalitis.

Eighteen of 298 immunized patients developed postimmunization aseptic meningoencephalitis (6%; 95% CI = 3.3 to 8.7%). This proportion is significantly higher than in the placebo group (0/74; Fisher's exact test, $p = 0.03$). There was no difference in the frequency of meningoencephalitis between Europe (12/160 exposed) and the USA (6/138) with stratified randomization ($\chi^2 = 1.30$, $p = 0.25$) and between France (6/97), other Europe (6/103), and the USA (6/172) when all patients enrolled were included ($\chi^2 = 1.28$, $p = 0.25$). Most of the cases diagnosed with meningoencephalitis presented with progressively increased confusion, headache, or lethargy. Other symptoms have been varied but consistent with possible meningitis and encephalitis, for example, fever, nausea, vomiting, seizures, drowsiness, disorientation, ataxia, difficult walking, decreased alertness, hemiparesis, and aphasia or speechlessness (see the case vignettes and table 1). The course was monophasic in most cases; four patients experienced relapses, which were severe in two cases.

CSF was studied in 17 of the 18 cases. With the exception of Patient 14, who had a CSF white blood cell count of

Table 1 Patient characteristics

Patient (country)	Age, y	Gender	No. of immunizations	Latency from last injection to symptom onset, d	Concomitant medications	Clinical summary
1 (F)	65	M	2	8	Donepezil, paroxetine, vitamin E, influenza virus vaccine	Mild meningoencephalitis with single cerebral lesion
2 (F)	67	M	1	16	Rivastigmine, trimetazidine, hydroxyzine, loratidine	Severe meningoencephalitis with multiple extensive brain lesions and severe relapse (see text)
3 (F)	61	M	2	13	None	Meningoencephalitis with multiple cerebellar and frontal lesions and transient relapse
4 (USA)	84	F	2	36	Donepezil, vitamin E, aspirin, conjugated estrogens, levothyroxine	Worsening of confusion, aphasia, retropulsion and loss of autonomy; mild CSF reaction; no MRI (refusal)
5 (F)	81	M	2	23	Donepezil, aspirin, zopiclone, bisoprolol/hydrochlorothiazide, hydroxyzine	Meningoencephalitis with single cerebellar lesion
6 (F)	77	F	2	30	Donepezil, paroxetine	Meningoencephalitis with single cerebral lesion plus widespread leukoencephalopathy
7 (USA)	83	F	3	5	Galantamine, paroxetine, vitamin E, calcium with vitamin D, aspirin, clopidogrel, digoxin, lansoprazole	Mild meningoencephalitis with single cerebral lesion (see text)
8 (USA)	81	F	2	51	Donepezil, citalopram, metoprolol, lisinopril, chlorazepate, hydrochlorothiazide	Clinical, CSF, and MRI features of meningitis with no focal neurologic signs
9 (USA)	80	F	2	42	Donepezil, conjugated estrogens, benazepril, levothyroxine, indapamide	CSF and MRI evidence of mild aseptic meningitis; no focal neurologic signs
10 (USA)	72	F	2	69	Donepezil, paroxetine, conjugated estrogens, loratidine, ephedrine	Clinical and CSF evidence of mild aseptic meningitis; no focal neurologic signs
11 (Eur)	75	M	2	57	Galantamine, tamsulosin, doxycycline	Mild meningoencephalitis with pontomesencephalic syndrome and bilateral cortical lesions
12 (F)	77	F	2	29	Donepezil, nitrendipine	Severe meningoencephalitis with bilateral basal ganglia lesions plus widespread small abnormalities
13 (Eur)	73	F	2	56	Lorazepam	Meningoencephalitis with single large deep temporal lesion
14 (Eur)	61	M	2	66	Galantamine	Mild meningoencephalitis with multiple bilateral cerebral lesions
15 (Eur)	75	F	2	52	Donepezil, paroxetine, aspirin, influenza virus vaccine	Meningoencephalitis with bilateral cerebral lesions; probable relapse with new lesion in left temporal lobe
16 (USA)	79	F	2	71	Donepezil, sertraline, vitamin E, vitamin C	Mild meningoencephalitis (no CSF study) with few discrete cerebral lesions, predominantly right temporal
17 (Eur)	69	F	2	156	Galantamine, sertraline, aspirin, estradiol	Mild meningoencephalitis without focal neurologic or MRI signs
18 (Eur)	76	M	2	168	Tamsulosin, aminoacetic acid, glutamic acid, buflomedil	Moderate meningoencephalitis with neurologic signs and bilateral white matter lesions predominantly in right hemisphere

F = France; Eur = other European countries.

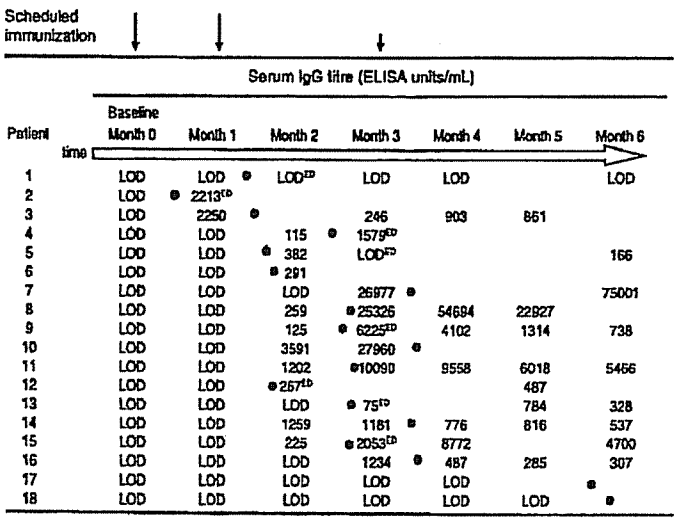

Figure 1. Schematic representation of serum antibody titers in temporal relation to scheduled immunizations and the time to onset of symptoms. With the exception of Patient 2 (one dose) and Patient 7 (three doses), all patients received two immunizations with AN1792 plus QS-21 as adjuvant before dosing was discontinued. No case occurred later than 6 months after the last immunization. Filled circles = experience onset; LOD = below lower assay limit of detection (50 ELISA U/mL); ED = sample taken at early discontinuation visit, not at monthly visit; Month 3: only one patient (Case 7) received a third injection.

3 cells/μL. all 17 had mononuclear pleocytosis (71 to 100% lymphocytes) with an initial white blood cell count that ranged from 15 to 130 cells/μL. Maximum protein content was between 0.33 and 3.1 g/L. Glucose level was between 3.3 and 5.4 mmol/L. With the exception of one patient, no oligoclonal bands were initially reported at electrophoresis; another case had oligoclonal bands at a later time. Virology was negative in most and slightly positive for herpes simplex virus (HSV) 1 in only one case. No bacteria were found in any case. IgG content was markedly elevated in three of four patients in whom it was tested, up to 98 mg/L.

ELISA titrations of anti-Aβ antibodies in CSF were performed in a subset of this meningoencephalitis cohort. Of the symptomatic patients, five have only baseline, four have only follow-up samples, and four have both. None of the baseline data are positive. The results for the follow-up cases show that six of the eight tested have elevated anti-AN1792 IgG titers and four have elevated IgM titers in CSF samples.

The cases have varied substantially in severity: Twelve patients have recovered to or close to their baseline status, whereas six have persistent disabling cognitive worsening, two of whom also have focal neurologic sequelae, extensive and severe in one case (table 2). All of these patients remain alive at the time of this report. The MRI findings with and without contrast have been variable: A few patients showed only meningeal enhancement, whereas others had meningeal thickening, white matter lesions, with or without enhancing or edema, and a majority had posterior cerebral cortical or cerebellar lesions (see table 2). None of the cases had a hemorrhagic component visible at MRI. With the exception of Patients 2 and 7, who received one and three immunizations, patients who experienced SAE received two doses of the study drug before symptom onset. Most patients were initially treated with antiviral therapy and antibacterials until an infectious etiology was ruled out. Some patients also received IV corticosteroids empirically, at variable doses and duration, the response to which was also variable but resulted in some level of improvement in most. Two patients (Patients 2 and 3) with severe signs and symptoms who did not respond to corticosteroid therapy received plasmapheresis. Patient 3 was reported as definitely having a positive response, whereas Patient 2 may have had a modest response, but this was not clear even after seven runs of plasmapheresis.

Analysis of serum samples by ELISA indicated that 15 of the 18 patients experiencing meningoencephalitis had antibodies against AN1792 (see figure 1). Serum IgG was not detectable in Patients 1, 17, and 18, although Patient 1 had a measurable IgM titer at the early discontinuation visit. All serum IgM levels in the meningoencephalitis patients were positive, though some are only minimally so.

Case vignettes. The case histories of two patients representative of the spectrum of clinical, brain imaging, and CSF manifestations encountered in the series are described in detail below. These and the other reported cases are summarized in tables 1 and 2, in chronologic order of reporting.

Patient 2. A 67-year-old man with mild AD (MMSE = 21) presented 16 days after the first injection of AN1792 with a 2-day history of frontal headache, nausea, and vomiting, but no fever. He developed weakness of the left leg and difficulty with balance and gait. The weakness progressed to involve the left arm, and he was admitted to the hospital the following day with a left hemiparesis and temporal–spatial disorientation. Two days later, he developed a right facial paralysis and lost the ability to swallow. He became less responsive but remained afebrile. CSF showed a marked elevation of protein (1.2 g/L), with normal glucose level and a white cell count of 165/μL (95% lymphocytes). A CT scan on day 3 revealed widespread subcortical periventricular hypodensity of the white matter, cortical atrophy, and ventricular enlargement. MRI showed widespread subcortical hyperintensities on fluid-attenuated inversion recovery (FLAIR) sequences in both hemispheres (figure 2A). Acyclovir therapy was initiated because of a positive test (later proven false) for HSV in the CSF. By

Table 2 Summary of clinical signs and MRI scans in patients experiencing encephalitis

Patient no.	MMSE score		MRI scan results. Days: from last injection	Fever	Clinical outcome
	Baseline	Lowest			
1	15	5	Day 51: probable area of demyelination in left parieto-occipital position	38.5–39 °C	Complete recovery
2	21	17	See legend to figure 2	No	Relapsed: severely dependent with prolonged hospitalization
3	26	17	Day 22: hypersignal in fossa posterior, vermis, lower cerebellar hemispheres, and right nucleus accumbens: T1 with gadolinium: billhook-like image, deep gyri between F1 and F2 in frontal and superior area, consistent with arterial or venous occlusion + 20 days: right temporal and bilateral occipital areas of hypersignal	No	Relapsed: complete recovery
4	19	"Very low" (no score)	Not obtainable (withdrawn from follow-up)	No	Partial recovery: neurologic and cognitive sequelae
5	26	19	Day 49: widespread cerebellar edema	37.5–38.3 °C	Complete recovery
6	24	6	Day 39: left frontal and right temporo-occipital cortical hypersignals and associated right temporo-occipital subcortical edema + 1 mo: Resolution of right temporo-occipital edema and no new lesions + 3 mo: Significant regression of previously described lesions + 5 mo: Almost complete regression of corticosubcortical contrast enhancement	No	Complete recovery
7	19	12	Day 9: heterogeneous area of abnormal signal involving right occipital lobe, considered most likely to represent subacute infarct: no meningeal enhancement	37.7–39.4 °C	Complete recovery
8	16	0	Day 76: abnormal meningeal enhancement and meningeal flare, mild ventricular enlargement, moderate small vessel ischemic changes in periventricular and deep cortical white matter	No	Partial recovery: cognitive sequelae
9	25	No data	Day 59: slight meningeal enhancement	Yes	Complete recovery
10	15	7	Day 77: cerebral atrophy with ventricular dilatation and cortical atrophy representing old ischemic changes or midline shifts: no acute lesion observed: impression was cerebral atrophy	Yes	Complete recovery
11	24	9	Day 57: evidence of older vascular lesions but no indexes of recent ischemia; increased leptomeningeal accumulation of contrast medium and bilateral parietocortical hyperintensities with suspicion of meningoencephalitis + 5 days: no new lesions, leptomeningeal enhancement markedly reduced	No	Complete recovery
12	16	No data	None reported	38 °C	Complete recovery
13	20	9	Day 69: extensive signal abnormality in right temporal lobe white matter, without enhancement, increasing 1 wk later + 2 mo: almost complete resolution of abnormalities	No	Partially recovered: cognitive sequelae
14	23	21	Day 66: cortical and subcortical hyperdensities in right and left occipital regions and frontal region with moderate local edema: left frontal and bilateral occipital leptomeningeal enhancement + 1 mo: marked improvement of all above abnormalities	No	Complete recovery
15	20	13	Day 64: abnormal signal intensities on both parieto-occipital regions, more important on left hemisphere, compatible with meningoencephalitis; diffuse leukoencephalopathy; corticosubcortical atrophy	No	Partial recovery: cognitive sequelae

Table continues

Table 2 Continued

Patient no.	MMSE score: Baseline	MMSE score: Lowest	MRI scan results. Days: from last injection	Fever	Clinical outcome
16	26	None reported	Day 100: abnormal lesion involving right posterior parietal and right occipital lobes with significant finger-like vasogenic edema with extension into temporal lobe; anterior displacement of left occipital horn, no significant gadolinium uptake; local brain swelling with effacement of local cortical sulci – 2 mo: mild cortical atrophy; almost complete resolution of right posterior parieto-occipital lobe lesion; no mass effect; increased signal intensity in left occipital pole and posterior parietal lobe, compatible with inflammatory changes	No	Complete recovery
17	25	15	Day 159: marked atrophy, meningeal enhancement of parieto-occipital region of left side At 3 mo (relapse): bilateral occipital meningeal and cortical enhancement	No	Relapsed; close to complete recovery after second episode
18	20	0	Day 297: bilateral lesions on white matter without enhancement after gadolinium with right hemispheric predominance – 20 days: improvement of inflammatory lesions and reduced white matter lesions – 1.5 mo (relapse): severe worsening of encephalitis lesions with cerebral edema	No	Relapsed; partial recovery; severe cognitive and functional sequelae

The time between onset of symptoms and MRI scanning is indicated.

MMSE = Mini-Mental State Examination.

day 5, the patient still had reduced consciousness and a left spastic hemiparesis. He was diagnosed as having acute disseminated encephalitis, probably postimmunization. On day 7, cognitive function became profoundly decreased, and the patient became mute and bedridden. Anti-AN1792 serum IgG measured at the onset was at 2,213 ELISA U. A repeat MRI scan was consistent with meningoencephalitis, including cerebellitis (see figure 2B). On day 16, the patient developed acute dyspnea with left pulmonary atelectasis, hypoxia, and hypocapnia. Antimicrobial therapy was initiated, together with a 4-day regimen of high-dose (1 g/day) IV methylprednisolone (MP) when HSV infection had been ruled out. By the second day of steroid therapy, the patient awakened, although the left-sided motor deficit persisted. An additional 3-day course of IV MP was begun on day 29, but there was no further improvement and his diabetes became severely decompensated. His neurologic status worsened around day 52 with new bilateral cortical–subcortical lesions on MRI, predominantly in the right frontal lobe and the left temporal lobe (see figure 2C). After a cerebral sinus thrombosis was ruled out by MRI, a series of plasmapheresis runs was started, with a total of seven treatments over 2 weeks. The patient's level of consciousness continued to fluctuate, and akinesia with left hemiplegia persisted. On day 74, the patient was transferred to a rehabilitation ward with a persistent severe left hemiplegia, akinesia, and loss of speech. At the time of this report, >12 months after the initial onset of symptoms, he remains hospitalized and totally dependent, although his level of consciousness and language continue to improve slowly. His most recent MRI showed bilateral widespread supratentorial white matter lesions (see figure 2D).

Patient 7. An 83-year-old woman presented with fever 5 days after receiving her third dose of AN1792. The following day, she awoke extremely confused, shivering, and akinetic, but without headache, visual disturbances, or focal weakness. She was admitted to the hospital where a diagnosis of community-acquired pneumonia was considered, and treatment with gatifloxacin was initiated. By the third day after presentation, the patient still had a mild fever, although she was more alert, coherent, and closer to baseline. MRI performed on day 4 showed a right cortical–subcortical abnormality, suggesting a subacute infarction. CSF findings indicated mild meningitis, and tests for HSV-1 and -2 were negative. On day 8, the patient was afebrile, and a clinical diagnosis of encephalopathy related to aseptic meningitis was made. The patient was still lethargic, speechless, and unable to follow commands on day 10. Four weeks after the initial presentation, she was alert and oriented to time, place, and person, walked with assistance, and had a mild receptive aphasia. On day 35, she began experiencing hallucinations, which continued for several days. At a follow-up visit 123 days after symptom onset, she had become clinically stable. In a later follow-up, she had recovered to her baseline condition.

Discussion. The subacute aseptic meningoencephalitis described in this report, which prompted cessation of the active dosing of AN1792, is probably a

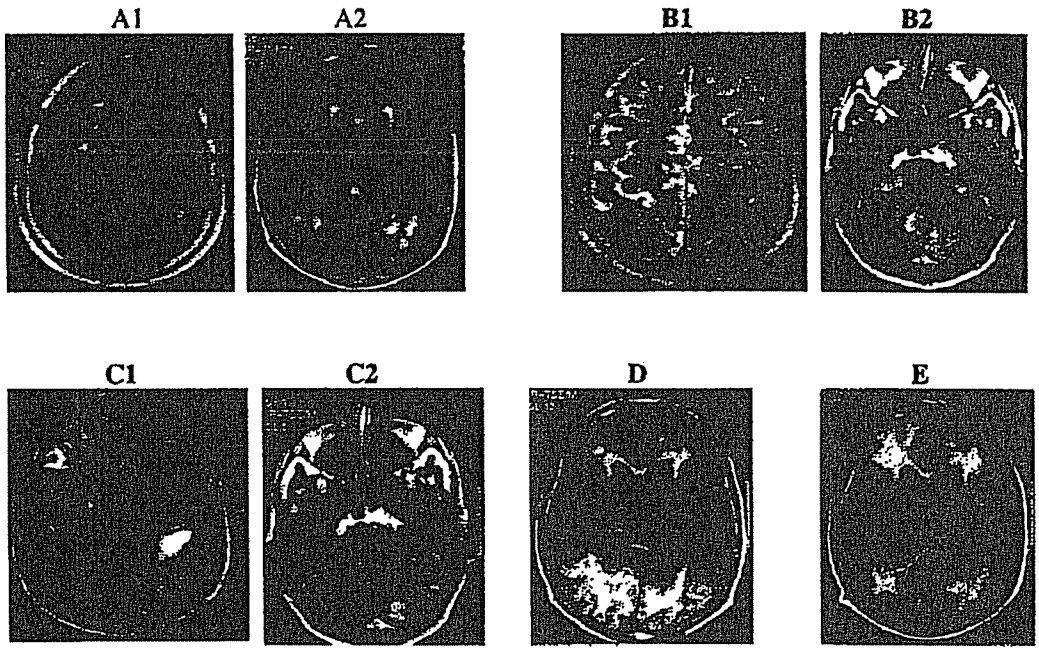

Figure 2. Serial brain MRI scans of Patient 2. A set of fast fluid-attenuated inversion recovery (FLAIR; repetition time = 11,000 milliseconds, echo time = 140 milliseconds, inversion time = 2,500 milliseconds) T2-weighted images performed 22 days (A1 and A2), 41 days (B1 and B2), 64 days (C1 and C2), 87 days (D), and 170 days (E) after immunization. (A) Presence of high signal intensities in the subcortical white matter and in the central sulcus (A1); numerous high signal intensities in the deep white matter (A2). (B) All sulci present an increased signal intensity that is related to the presence of protein in the CSF (B1); new lesions in the white matter of the right cerebellar peduncle (B2). (C) Worsening in the number and location of the lesions, now affecting the white matter and the gray matter of the cerebral (C1) and the cerebellar cortex (C2), while the lesions in the cerebellar peduncle have disappeared. (D) Extensive new lesions in the deep posterior white matter also affecting the adjacent cortex. (E) New lesions in the deep frontal white matter, while some lesions in the posterior white matter disappeared. (Courtesy of Prof. Vincent Dousset, trial neuroradiologist).

side effect of active anti-Aβ immunization, as it occurred only in patients who received active drug and meningoencephalitis is not a known complication of AD itself. This complication was unexpected, as it had not been observed in the preclinical studies or in the Phase I clinical trials in which AN1792 and QS-21 were administered alone and in combination.[11] It occurred mostly (16/18 patients) within 3 months after the first injection and not later than 6 months after the last one. It was more often self-limiting within a few weeks, but relapses occurred in four patients within months and severe sequelae persisted in six after 6 to 12 months of follow-up.

Meningoencephalitis occurred in 16 of the 18 cases after two injections, after one injection in one (Patient 2, the most severely affected patient), and after three injections in one case (Patient 7, who had a favorable outcome). Thus, there is no obvious relationship between the number of injections and the severity of the postvaccination meningoencephalitis. Serum IgG against AN1792 was not detectable in three patients, and there was no consistent correlation of antibody titers with the delay to symptom onset, the severity of encephalitis, or the occurrence of relapses. None of these cases had any hemorrhagic component at brain imaging, in contrast to a report of increased cerebral microhemorrhage and hematomas in APP23 transgenic mice with cerebral amyloid angiopathy (CAA) passively immunized against human Aβ,[15] a model naturally prone to cerebral hemorrhage. Microhemorrhages and small hematomas like those reported in this mouse model may not have been large enough to be detected by MRI in our patients. However, only three patients of 372 in the whole trial experienced a cerebral hemorrhage during the course of the study: one under placebo and two under active treatment (one hemorrhagic infarction and one possibly of the CAA type). These two cases had no features in common with the complication described in this article.

This postvaccination meningoencephalitis series has several features in common with the rare syndrome of delayed postvaccination meningoencephalomyelitis,[16] which occurs in 1 of 1,000,000 measles

vaccinations,[17] and the more frequent postinfection acute disseminated encephalomyelitis (ADEM),[18,19] which occurs in about 1 of 1,000 cases of measles.[20] The clinical presentation of some of our cases is similar to that of ADEM, with the acute or subacute onset of fever with symptoms and signs of diffuse or multifocal CNS involvement. ADEM consists of a monophasic, self-limiting course in a majority of cases but sometimes follows a relapsing course,[21] as in our Patients 2, 3, 17, and 18. For some authors, MRI abnormalities are mandatory for the diagnosis of ADEM[22] or are consistently found,[18] whereas for others, they are not.[23] When MRI abnormalities are present, some of the parenchymal MRI aspects of ADEM are similar to those of our cases.[22,24]

The syndrome reported here, however, differs from ADEM in several respects. First, ADEM was described mainly in children[22,24] and young adults,[18,23,25] whereas the patients in this series were from 61 to 84 years old (see table 1). Second, the syndrome reported here occurred with a median delay of 75 days (minimum of 16 days) after the first immunization, the two most delayed cases as late as 6 and 6.5 months, in contrast to the typical 6- to 15-day onset of ADEM after an infectious episode or vaccination.[23] Third, there was no clinical or MRI evidence of myelitis in our cases. In addition, cranial nerve palsies and optic neuritis frequently occur in ADEM,[23,25] but they did not appear in any of our cases. Finally, the almost consistent CSF meningeal reaction in our cases was absent in half the reported cases of ADEM.[18,23] Nevertheless, ADEM is a heterogeneous disorder, and some of the cases described as such in the literature in older adults are similar to those described here.

At this stage, we can only speculate about the role of abnormal cellular or humoral immunoreactions in the pathogenesis of the postvaccination CNS reaction associated with Aβ immunization. The antibodies generated in the blood of vaccinated patients showed a high specificity for the Aβ in the plaques and blood vessels of the brain.[26] There was neither a cross-reaction with endogenous Aβ-protein precursor and its derivatives nor with normal brain cells.

Serum antibodies against Aβ were detectable in 15 of the 18 cases, far in excess of the 25% expected from the previous Phase I trial for all patients exposed. CSF antibodies were present in six of eight patients tested after onset of encephalitis. So there is an obvious relation between the presence of the antibodies and the risk of encephalitis. But as there was no apparent correlation with the titers of serum anti-Aβ42 antibody and either delay, severity, or occurrence of relapses, a potential inflammatory mechanism mediated by activated T-cells is currently being debated,[27] as it was shown that the Aβ42 molecule contains a T-cell-activating domain.[28] Indeed, in the only pathologic case so far, recently reported from the previous Phase I study,[29] postmortem examination of the brain from one patient who received AN1792 showed a widespread T-lymphocyte meningoencephalitis likely to correspond to the side effect described clinically in the patients in this study. That patient also had extensive infiltration of cerebral white matter by macrophages and evidence to suggest that the immune response had elicited clearance of Aβ plaques. This issue may be clarified in the current series when the analysis of the full data set will allow comparison of humoral and cellular immunity responses between cases and noncases.

Another possibility is that inflammatory signs may occur during the clearance of amyloid from the brain as part of the intended therapeutic process. Indeed, Aβ immunization was associated with a transient increase in microglial activation in transgenic mice vaccinated monthly for 3 to 5 months.[30] Remarkably, microglial activation had vanished after 9 months of treatment. In our series, no case occurred later than 6 months after the first injection or 5 months after the last (see figure 1). Thus, it is possible that Aβ immunization and Aβ clearance from the brain are concomitant with transient periods of inflammation.

In addition, some of the Aβ deposited in AD is located in brain blood vessels, and amyloid-laden vessels may be included in the amyloid clearance process, thereby disrupting blood vessel integrity and allowing a leak of proteins through the blood–brain barrier and possibly microhemorrhages. The frequent posterior cortical involvement at MRI in patients with postvaccination meningoencephalitis in our cases (see table 2) might be explained by the abundance of parenchymal β-amyloid deposits in the posterior cerebral[31] and cerebellar[32] cortices in advanced AD. It is not clear, however, why the brain areas most affected by amyloid deposition, namely, the temporal and associative cortices,[33] were not predominantly or even consistently affected by the putative anti-Aβ autoimmune process.

The findings reported from this study represent analysis of only those patients who experienced a serious CNS AE consistent with the diagnosis of meningoencephalitis. Although dosing and follow-up were terminated after the fourth report, the study remains unblinded until all data are checked and the primary statistical analyses are completed. The analysis of the full study results, particularly the immunologic data, will yield additional clues in interpreting the observations reported here, as well as in obtaining information on the effect of AN1792 on efficacy endpoints, including MRI and cognitive parameters.

Future immunotherapeutic strategies may consider active immunization with immunoconjugates composed of parts of the Aβ molecule, specifically excluding the epitope that may provoke abnormal T-cell reactions.[11] Therapeutically effective antibodies targeting Aβ residues 4 to 10 in mice can inhibit cytotoxicity and fibrillogenesis in cellular models.[34] In addition, passive immunization strategies with humanized anti-Aβ antibodies are currently being developed. Information on safety and pilot efficacy

collected from this and other immunization trials will be of crucial value for the future development of safe and effective immunotherapies for AD.

Note added in proof. Since the manuscript was revised, one death was reported, on March 9, 2003 (Case 18), as a sudden death probably associated with inhalation, several months after the encephalitis.

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